to changes in transmural pressure by active changes in its diameter in the sense of Bayliss response. This myogenic mechanism very likely contributes to the postnatal closure of DA as a factor potentiating responses to increase in  $pO_2^3$ , and in extreme situations (e.g. fetal asphyxia) or in certain species with weakly reacting DAs to oxygen<sup>8,10</sup>, it may even play the role of the main factor.

The observed sudden dilation of DA, associated with depression in its responsiveness when pressures 90 mm Hg-130 MM Hg are reached, may represent one of several pathogenic factors causing ductus arteriosus apertus. Such a dilation may be due to changes in histoarchitectonics<sup>11</sup>, or ultrastructure in DA wall resulting from its distension to a critical degree.

## Possible significance of aminotransferases in tissues of the aestivating fresh water mussel, *Lamellidens marginalis* (Lamarck)

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Summary. Both aspartate and alanine aminotransferase levels increased in digestive gland, foot and mantle on aestivation. The free amino acids and pyruvic acid also increased in all tissues. The significance of these changes is discussed in relation to gluconeogenesis.

Some species of fresh water gastropods<sup>2-5</sup> and pelecypodes<sup>6</sup> are known to aestivate under drought conditions. Studies on aestivation metabolism, mostly in gastropods, showed decrease in succinate dehydrogenase, cytochrome, C oxidase<sup>7,8</sup> and acetylcholine esterase<sup>9</sup>.

Since the animals during aestivation desists from feeding and mainly depend on glycogen reserves, it is obvious that these reserves also deplete in course of time<sup>2,5</sup>. This necessiates the aestivating animal to depend on other sources for its survival. It is viewed that aminotransferase enzymes namely aspartate aminotransferase EC.2.6.1.1 (AAT) and alanine aminotransferase EC.2.6.1.2 (AlAT) to serve as link enzymes for carbohydrate and protein metabolisms<sup>10</sup>. Since these enzymes interconvert strategic compounds and act as sources for keto acids for Krebs' cycle and for gluconeogenesis 10, in the present study these enzymes were selected to ascertain its role during aestivation of fresh water mussel. Since the literature on aestivation in fresh water mussel is scanty, and no work has been done in fresh water mussels on these lines, an attempt has been made to correlate aminotransferase activity pattern with the aestivation phenomenon.

Materials and methods. The fresh water mussel (Lamellidens marginalis (Lamarck)) were collected from the ponds and adapted to laboratory conditions. They were fed with fresh water plankton. Aestivation was induced by keeping at a time 50 specimens in a glass troughs containing soil from the collecting spot and covered with little water. The water was allowed to evaporate at room temperature as suggested by Newman and Thomas<sup>4</sup>. Only 1 month aestivating specimens were selected for experimentation.

The AAT and AlAT activity levels in selected tissues were estimated by the method of Reitman and Frankel<sup>11</sup> after standardization. 3 tissues, viz. digestive gland, foot and mantle, were excised and homogenized in cold 0.25 M sucrose solution using Yorco tissue homogenizer (Yarco Scientific Industries, New Delhi) and centrifuged at 600× g. The supernatant was used for assay. The reaction mixture consists of 100 µmoles phosphate buffer (pH 7.4) 2 μmoles of a-oxoglutaric acid (pH 7.4) 20 μmoles of Laspartate (pH 7.4) (for AAT); 50 µmoles of DL-alanine (pH 7.4) (for AlAT) and 0.1 ml of supernatant (10% w/v) as enzyme source. After incubating for 30 min, the reaction was arrested by adding 1 ml of 2,4 dinitrophenyl hydrazine (0.001 M in 0.1 N HCl). After 20 min, the colour was developed by adding 10 ml of 0.4 N sodium hydroxide solution. The colour was read at 546 nm in spectrophotometer model-CL-20 (Elico, India). The protein content was determined by using Folin Ciocalteu's reagent<sup>12</sup>. The free amino acid content was estimated by the method of

Levels of aspartate and alanine aminotransferases and organic acid. (Free amino acid and pyruvic acid) content in selected tissues of aestivating fresh water mussel. Each value is a mean of 6 individual observations  $\pm$  SD

Enzyme/organic acids	Digestive glar Control	nd Aestivated	Foot Control	Aestivated	Mantle Control	Aestivated
Aspartate aminotransferase*	5.075 ± 0.1766	6.925 ± 0.1893 + 36.45 p<0.001	3.01 ± 0.1546	5.05 ± 0.225 + 67.74 p < 0.001	6.45 ± 0.447	7.0 ± 0.208 + 8.65 p < 0.02
Alanine aminotransferase*	5.04 ± 0.171	7.015 ± 0.1995 + 39.18	$\begin{array}{c} 3.62 \\ \pm  0.1393 \end{array}$	4.82 ± 0.224 + 33.14	$\pm 0.2316$	7.56 ± 0.2454 + 23.4
Free amino acids**	866.6 ± 30.76	p<0.001 1179.3 ± 46.05 + 33 p<0.001	$1007.83 \pm 25.16$	$p < 0.001$ $1198$ $\pm 27.2$ $+ 18$ $p < 0.001$	390.3 ± 26.5	p<0.001 434.3 ± 24.46 + 11.5
Pyruvic acid***	0.1007 ± 0.0047	$ \begin{array}{r}     0.125 \\     \pm 0.0056 \\     + 24.13 \\     p < 0.001 \end{array} $	0.0412 ± 0.001509	0.0662 ± 0.006 + 60.67 p<0.001	$\begin{array}{c} 0.0309 \\ \pm 0.0024 \end{array}$	$\begin{array}{c} p < 0.001 \\ 0.056 \\ \pm 0.0029 \\ + 81.2 \\ p < 0.001 \end{array}$

<sup>\*</sup> Values expressed as µmoles pyruvate/mg protein/h. \*\*Values expressed as µmoles tyrosine/g wet weight of tissue. \*\*\* Values expressed as mg pyruvic acid/g wet weight of tissue. +, indicates percent increase over control. p=t-test; all values are found to be significant.

Moore and Stein<sup>13</sup>. The pyruvic acid was estimated by the method of Friedman and Haugen<sup>14</sup>.

Results and discussion. The AAT and AlAT enzymes were found to increase in all tissues of the aestivating mussel. The increase in amino-transferases is in consonance with the elevation in free amino acids content (table). Since the aminotransferases are known to convert stategic compounds, like a-ketoglutarate, pyruvate, oxaloacetate, glutamate alanine and aspartate, and function as link enzymes between carbohydrate and protein metabolisms by acting as sources for ketoacids and for gluconeogenesis10, the same could be envisaged of the role of aminotransferases during aestivation of mussel.

During aestivation due to lack of feeding, the animal depends on its glycogen reserves. Previous reports showed depletion of glycogen in the aestivating gastropods<sup>2,3</sup> and the same trend was observed in the aestivating fresh water mussel<sup>15</sup>. Hence it is likely that, in order to meet the energy demands, the aestivating mussel depends on other sources for its survival. Since the free amino acid content increases in tissues of the aestivating animal; this serves as a precursors for aminotransferases, by converting strategic compounds 10 to meet the energy demands during the torpid stage. Since the activities of aminotransferases are known to alter under physiological conditions<sup>16</sup>, the increase in aminotransferases activity in all tissues is justifiable.

On aestivation the elevation of AAT is more heterogeneous with foot showing maximal activity (67.74%) while the elevation of AlAT in all tissues is more or less homogeneous, i.e. the activity level in all the tissues ranges from 23 to 39% (table).

The AAT/AlAT ratios were found to alter in the tissues of aestivating mussel. The AAT/AlAT ratios of foot was 1.0 while mantle and the digestive gland were 0.9 and 0.95 respectively. This shows that the mantle and digestive gland are more prone towards AlAT while the foot is relatively

more prone towards AAT. In general the AAT/AlAT ratios suggests a shift in enzyme activity pattern, showing preponderance towards AlAT enzyme activity which is in agreement with the increase in pyruvic acid content in all tissues of the aestivating mussel (table). Hence it can be reasonably inferred that the over-all increase in aminotransferases, particularly AlAT enzyme, may contribute effectively for gluconeogenesis to meet the energy demands during the period of suspended animation.

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Mise en évidence de l'excrétion urinaire d'antigènes de la membrane basale glomerulaire (MBG) dans l'urine de 3 rongeurs (rat, souris et cobaye)

Evidence of urinary glomerular basement membrane (GBM) antigens excretion in 3 rodents (rat, mouse and guinea-pig)

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Summary. Immunodiffusion technique, with rabbits antibodies against rat glomerular basement membrane (GBM), permits to evidence the presence of GBM antigens into normal urines of 3 rodents (rat, mouse and guinea-pig). These results confirm the earlier works in human and rabbit urines and show the antigenic communauty existing between the 3 rodents GBM, since we can evidence antigens of 3 different mammals with the same antibody. The origin and the nature of this patterns and the signification of this presence into mammalian urines are discussed.

Des produits antigéniques solubles issus de la membrane basale glomérulaire (MBG) ont été mis en évidence dans l'urine de certains mammifères, notamment chez l'homme<sup>4-6</sup>, chez le lapin<sup>7-9</sup> et très récemment chez le rat<sup>10</sup>. La présence éventuelle de tels produits est recherchée dans l'urine concentrée de souris et de cobaye par la méthode d'immunodiffusion<sup>11</sup> à l'aide d'anticorps de lapin anti-MBG de rat.

Matériel et méthodes. Préparation des MBG. La pulpe corticale de 200 reins de rat est broyée au moyen d'une spatule sur des tamis aux pores de 74 µm de côté. Les glomérules traversant ce premier tamis sont recueillis sur un second aux pores plus petits (53 µm de côté). La suspension de glomérules obtenue est abondamment lavée, puis traitée au sonicateur pendant 15 min. Des lavages successifs la débarrassent de tout élément cellulaire et permettent ainsi l'obtention d'une suspension de MBG pure qui est lyophilisée. Le contrôle de pureté de cette suspension est effectué en microscopie électronique.

Préparation des anticorps anti-MBG chez le lapin. 0,4 ml de la MBG brute est émulsionnée à volume égal par de l'adjuvant de Freund complet et injectée le 1er chaque lapin dans le coussinet plantaire. La même dose est administrée par voie intramusculaire après 3 semaines, tous les 8 jours pendant 6 semaines. Lorsque la présence d'anticorps spécifiques contre la MBG, solubilisée par 2 stérilisations à l'autoclave pendant 2 h<sup>12</sup> est nettement décelable dans le sérum des animaux par immunodiffusion, les animaux sont saignés et les sérums collectés sont réunis pour former un pool sérique d'anticorps anti-MBG.